

# Influence of environmental factors on after-ripened crowfootgrass (*Dactyloctenium aegyptium*) seed germination

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Laboratory and greenhouse studies were conducted to determine the effect of temperature, pH, water stress, and planting depth on crowfootgrass germination. When treated with constant temperature, crowfootgrass germinated over a range of 15 to 40 C, with the optimum germination occurring at 30 C (42%). Onset, rate, and total germination (94%) were greatest in an alternating 20 and 35 C temperature regime. Germination decreased as pH increased, with greatest germination occurring at pH 4 and 5. Germination was reduced when seed was subjected to water stress, and no germination occurred below  $-0.8$  mPa. Emergence was similar when seed were placed on the soil surface or buried at depths of 0.5 or 1 cm. Germination decreased with burial depth, and no seed emerged from 10 cm. These data suggest that crowfootgrass may emerge later in the season with warmer temperatures and after a precipitation event, and may emerge rapidly. These attributes could contribute to poor control later in the season by soil-applied herbicides or allow crowfootgrass to emerge after final postemergence treatments are made.

**Nomenclature:** Crowfootgrass, *Dactyloctenium aegyptium* (L.) Willd. DTTAE.

**Key words:** Alternating temperature, pH, scarification, water stress, weed biology.

The genus *Dactyloctenium* is widespread, and member species occur mostly in dry, sandy soils ranging in habitat from meso- to xerophytic (Holm et al. 1979; Watson and Dallwitz 1992). Crowfootgrass is a native of the old world tropics, and is among the 20 most globally widespread weeds (Holm et al. 1979; Simpson 1990). It occurs as both a weed and an important pasture species (Sharma and Chivinge 1982) and is the only representative species of the genus in the southeastern United States (Hitchcock and Agnes 1971; Radford et al. 1973). It is considered a weed in peanut production regions of the southeast United States, infesting 25 and 75% of the southeastern and Virginia–Carolina production regions, respectively (Bridges et al. 1994a, 1994b). Information about the germination and seedling establishment requirements is available for populations in Africa and India (Kumar et al. 1971; Okusanya and Sonaike 1991; Sharma and Chivinge 1982). The species has received little attention in the Americas apart from its inclusion in after-ripening studies (Taylorson and Brown 1977).

In previous experiments in India and the United States, crowfootgrass required either after-ripening treatments or a 5 mo or more period of storage for after-ripening (Gupta 1973; Taylorson and Brown 1977). Light was required for germination in several studies (Okusanya and Sonaike 1991; Taylorson and Brown 1977) but was not found to be a requirement by Kumar et al. (1971). Most research has focused on relieving the dormancy of crowfootgrass; however, several studies have examined depth of emergence and effects of substrate pH, salinity, and dry matter production (Adu et al. 1994; Gupta 1973, Kumar et al. 1971; Okusanya and Sonaike 1991; Sharma and Chivinge 1982).

Studies of germination and seedling establishment requirements yield basic ecological information for soil emergence (Bhowmik 1997). Such information can be used to

characterize the competitiveness and the potential infestation range of the weed as well as enhance management practices, allowing biological, chemical, or mechanical control options to be properly timed (Bhowmik 1997; Dyer 1995; Potter et al. 1984; Wilson 1988). Therefore, research was initiated to gain an understanding of the germination requirements of this problematic annual grass. The objectives of this research were to determine crowfootgrass germination response to temperature, solution pH, water stress, and planting depth.

## Methods and Materials

Crowfootgrass seed were harvested from fallow fields near Clayton, NC on September 3, 2000. Crowfootgrass seed were dormant at the time of collection, with  $5 \pm 2\%$  germination. The seed were allowed to dry to 11% moisture and stored at room temperature (28 C) for 6 mo before their use in experiments. The seed were sieved to remove any extraneous plant or floral material. The sieved seed were further cleaned in an air column separator<sup>1</sup> and tested for viability using 1% tetrazolium chloride solution before each trial (Peters 2000).

Previous reports (Okusanya and Sonaike 1991; Taylorson and Brown 1977) indicated crowfootgrass germination was dependent on light in germination chambers.<sup>2</sup> Therefore, light was provided for 8 h to coincide with the length of the high-temperature component of the temperature regime for all studies conducted in germination chambers. Studies on the gradient table were also provided with light for 8 h. Observations were made during the 8-h light period.

Crowfootgrass seed was  $93 \pm 4\%$  viable by tetrazolium chloride tests (Peters 2000) before each study was conducted (data not shown). Crowfootgrass seed required a consider-

able period of storage or after-ripening before dormancy was relieved (Gupta 1973). Consequently, seed were stored at room temperature (28 C) for a period of 6 mo before experimentation was begun. Germination before and after storage was  $9 \pm 4\%$  and  $91 \pm 4\%$ , respectively, in a 20 and 30 C regime. It was also necessary to remove the pericarp before germination experiments because greater germination was observed without it in place (Gupta 1973).

## Effect of Temperature

The effect of constant temperature was evaluated by evenly spacing 20 crowfootgrass seed in 25-ml Erlenmeyer flasks containing three pieces of filter paper<sup>3</sup> and 8 ml of deionized water. Experiments performed on the gradient table precluded randomization because the zones of temperature were fixed in position (Larsen 1965). The flasks were arranged on a thermogradient table (Larsen 1965) in six lanes corresponding to constant temperatures of 15, 20, 25, 30, 35, and 40 C, with six replicate flasks per temperature lane. Flasks were sealed using Parafilm to retain moisture. Light was provided by fluorescent overhead bulbs set for a 8 h light 16 h dark regime with a light intensity of  $30 \mu\text{mol m}^{-2} \text{s}^{-1}$ . Daily germination counts were made for the first 7 d, then every 3 d until no seed germination was observed for two observations. Each seedling was removed when a visible radicle could be discerned (Baskin and Baskin 1998). The study was conducted twice and the data combined for analysis.

A second study was conducted in growth chambers to determine crowfootgrass response to diurnal temperature. A randomized complete block design with four replications of treatments was used, and the study was conducted twice. Each replication was arranged on a different shelf within the respective germination chamber. Blocks were considered as replications over time. Twenty-five crowfootgrass seed were evenly spaced in 110-mm diam by 20-mm petri dishes containing two pieces of germination paper<sup>4</sup> and 10 ml of deionized water. Four temperature regimes were selected to reflect typical seasonal variation in North Carolina. The regimes 10 and 25 C, 15 and 30 C, 20 and 30 C, and 20 and 35 C, correspond to mean daily low and high temperatures for the months of May, June, July, and August, respectively, in Goldsboro, NC (Owenby and Ezell 1992). The high temperature component of the regime was maintained for 8 h. Light was provided by fluorescent overhead bulbs in 8 h light and 16 h dark with a light intensity of  $35 \mu\text{mol m}^{-2} \text{s}^{-1}$ . Light quality for the germination chamber is presented in Figure 1. Daily germination counts were made for 7 d, then every 3 d until no seed germination was observed for seven continuous days. Each seedling was removed upon germination as previously mentioned. The study was conducted twice and the data combined for analysis.

## Effect of Solution pH

A study with a randomized complete block design and four replications of treatments was used to examine the effects of pH on crowfootgrass germination. Each replication was arranged on a different shelf within the respective germination chamber. Blocks were considered study replication over time. Buffered solutions were prepared according to the

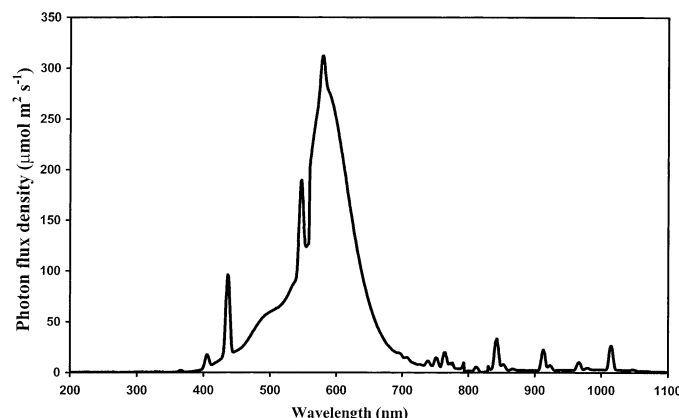


FIGURE 1. Light quality for germination chamber measured at 0.5 m from light source.

method described by Gortner (1949), using 0.1 M potassium hydrogen phthalate in combination with either 0.1 N HCl or 0.1 N NaOH to obtain solution pH values of 4, 5, and 6. A 25-mM sodium tetraborate decahydrate solution was used in combination with 0.1 N HCl or 0.1 N NaOH to prepare solutions with pH values of 7, 8, and 9. Twenty-five crowfootgrass seed were placed in petri dishes containing 10 ml of the buffer solution and the petri dishes placed in 10 and 25 C, 15 and 30 C, 20 and 30 C, and 20 and 35 C germination chambers. Germination was determined as previously mentioned. The study was conducted twice and the data combined for analysis.

## Effect of Water Stress

A study with a randomized complete block design and four replications of treatments was used to examine the effects of water stress on crowfootgrass germination. Each replication was arranged on a different shelf within the respective germination chamber. Blocks were considered study replication over time. Solutions with water potentials of 0.0, -0.3, -0.4, -0.6, -0.9, and -1.2 mPa were prepared by dissolving 0, 154, 191, 230, 297, and 350 g of polyethylene glycol<sup>5</sup> (PEG), respectively, in 1 L of deionized water (Michel 1983). Twenty-five crowfootgrass seed were placed in petri dishes containing 10 ml of PEG solution and the petri dishes placed in 10 and 25 C, 15 and 30 C, 20 and 30 C, and 20 and 35 C germination chambers. Germination was determined as previously mentioned. The study had four replications of treatments and was conducted twice and the data combined for analysis.

## Depth of Emergence

A depth of emergence study was conducted to examine the effect of burial depth on crowfootgrass seed emergence. The study design was a randomized complete block with treatments replicated four times in a glasshouse at an average daily temperature of  $33 \pm 5$  C and a nightly temperature of  $23 \pm 5$  C. Natural light supplemented with fluorescent lamps at a light intensity of  $300 \pm 20 \mu\text{mol m}^{-2} \text{s}^{-1}$  was used to extend the daylength to 14 h in the glasshouse study and to simulate field conditions in June.

Containers were filled to a depth of 10 cm with a Norfolk loamy sand soil (fine-loamy, siliceous, thermic, Typic Paleu-

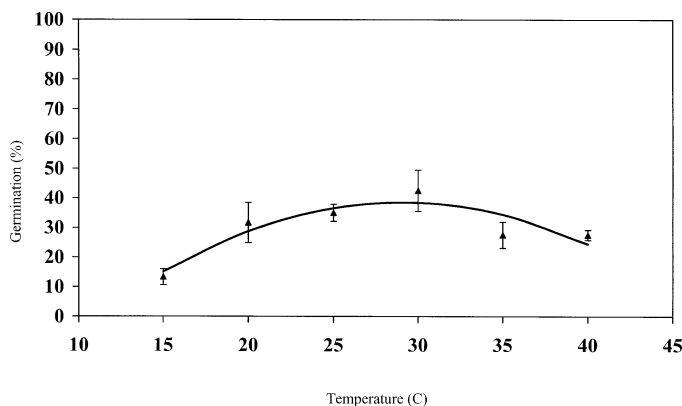


FIGURE 2. Influence of constant temperature on cumulative germination of crowfootgrass seed at 8 d as described by the equation  $y = -61.3 + 6.9(\text{temp}) - 0.12(\text{temp})^2$ ,  $R^2 = 0.81$ . Vertical bars represent standard errors of the mean.

dults). Containers were 15 cm (diameter) by 18 cm (height). Twenty crowfootgrass seed were placed on the soil surface or covered to depths of 0.5, 1, 2, 4, 6, or 10 cm with the same soil. Pots were subirrigated before planting to field capacity, and then surface irrigated daily to field capacity. Emergence counts were recorded daily for the first 7 d, then every 3 d until no seed germination was observed for seven continuous days. Plants were considered emerged when a cotyledon could be visibly discerned. The study was conducted three times and the data combined for analysis.

## Statistical Analysis

Data variance was visually inspected by plotting residuals to confirm homogeneity of variance before statistical analysis. Both nontransformed and arcsin-transformed data were examined, and transformation did not improve homogeneity. Analysis of variance (ANOVA) was therefore performed on nontransformed percent germination. Trial repetition and linear, quadratic, and higher order polynomial effects of percent germination over time were tested by partitioning sums of squares (Draper and Smith 1981). Regression analysis was performed when indicated by ANOVA. Nonlinear models were used if ANOVA indicated that higher order polynomial effects of percent germination were more significant than linear or quadratic estimates. Estimation used the Gauss-Newton algorithm, a nonlinear least squares technique (SAS 1998).

Germination resulting from constant temperature treatments was described by a parabolic model of the form:

$$y = \beta_0 + \beta_1 \text{temp} + \beta_2 \text{temp}^2 \quad [1]$$

where  $\beta_0$ ,  $\beta_1$ , and  $\beta_2$  are the intercept, first-, and second-order regression coefficients, respectively, and  $y$  is the cumulative germination at temperature "temp". A parabolic model was used to describe the germination of crowfootgrass because the constant temperature used in the experiment allowed direct correlation of germination response.

ANOVA indicated higher order polynomial effects for germination resulting from alternating temperature treatments, solution pH treatments, and water potential treatments. Thus, the germination response for each treatment was modeled using the logistic function:

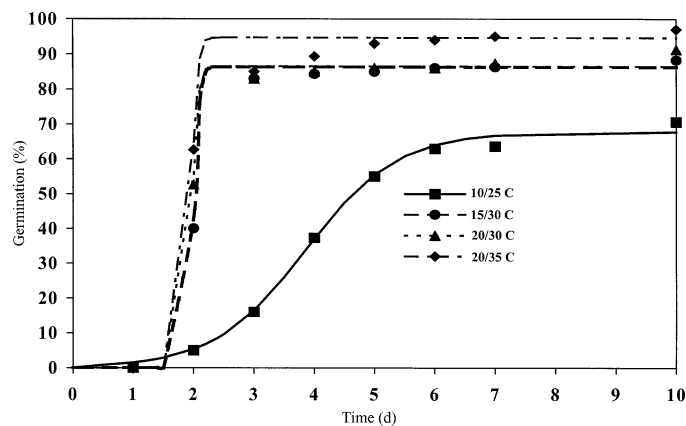


FIGURE 3. Influence of four temperature regimes on crowfootgrass germination, modeled using equation  $y = M (1 + \exp[-K(t - L)])^{-1}$  with estimated parameters (and standard errors) as follows:

Temperature	<i>M</i>	<i>K</i>	<i>L</i>	<i>R</i> <sup>2</sup>
10/18	67.8 (2.9)	1.32 (0.37)	3.9 (0.2)	0.83
15/30	86.3 (2.9)	20.40 (0.24)	2.0 (0.1)	0.81
20/30	86.6 (3.2)	19.65 (0.38)	2.0 (0.1)	0.77
20/35	94.7 (2.3)	19.51 (0.27)	2.0 (0.1)	0.88

$$y = M (1 + \exp[-K(t - L)])^{-1} \quad [2]$$

where  $y$  is the cumulative percentage germination at time  $t$ ,  $M$  is the asymptote or theoretical maximum for  $y$ ,  $L$  is the time scale constant or lag to onset of germination, and  $K$  is the rate of increase (Roché et al. 1997). Estimation used the Gauss-Newton algorithm, a nonlinear least squares technique (SAS 1998). When a nonlinear equation was fit to the data, an approximate  $R^2$  value was obtained by subtracting the ratio of the residual sum of squares to the corrected total sum of squares from one (Askew and Wilcut 2001; Draper and Smith 1981).

Depth of emergence data were subjected to an ANOVA using the general linear models procedure SAS (SAS 1998). No crowfootgrass plants emerged from 10 cm, and consequently these data were not included in the analysis. Sums of squares were partitioned to evaluate planting depth and trial repetition. Both study replication and repetition were considered random variables, and main effects and interactions were tested by the appropriate mean square associated with the random variable (McIntosh 1983).

## Results and Discussion

### Effect of Temperature

Crowfootgrass germination was influenced by temperature. When exposed to constant temperature, crowfootgrass seed germinated over a temperature range of 15 to 40 C (Figure 2). Constant temperature resulted in a maximum germination of 43% at 30 C. Our data support those reported by Gupta (1973) and Kumar et al. (1971), who reported an optimal constant temperature of 30 C for crowfootgrass seed germination.

ANOVA indicated a significant temperature regime by germination interaction, so crowfootgrass germination is presented for each temperature regime (Figure 3). Maximum cumulative germination (94%, parameter  $M$ ) of crowfoot-

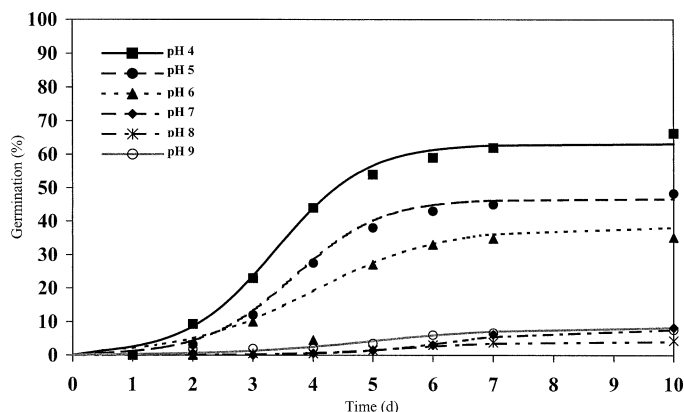


FIGURE 4. Influence of solution pH on crowfootgrass germination averaged across temperature regimes, modeled using the equation  $y = M(1 + \exp[-K(t - L)])^{-1}$  with estimated parameters (and standard errors) as follows:

pH	<i>M</i>	<i>K</i>	<i>L</i>	<i>R</i> <sup>2</sup>
4	63.1 (2.7)	1.34 (1.29)	3.4 (0.2)	0.99
5	46.7 (2.0)	1.37 (0.44)	3.7 (0.2)	0.99
6	38.2 (2.2)	0.95 (0.17)	4.0 (0.4)	0.99
7	8.6 (0.3)	1.21 (0.21)	6.3 (0.2)	0.99
8	4.1 (0.2)	1.2 (0.23)	5.5 (0.3)	0.99
9	8.3 (0.5)	0.88 (0.21)	5.0 (0.4)	0.99

grass occurred when seed were exposed to a 20 and 35 C regime. The germination rate (parameter *K*) in response to the 20 and 35 C regime was similar to the rates at 15 and 30 C and 20 and 30 C regimes. Time to 50% germination (parameter *L*) in the 15 and 30 C, 20 and 30 C, and 20 and 35 C temperature regimes was 2.0 d. Total percentage of cumulative germination was lowest in the 10 and 25 C regime. The lower cumulative germination at low temperature may indicate that crowfootgrass germinates in the warmer portion of the growing season which would include June, July, and August. Goosegrass [*Eleusine indica* (L.) Gaertn.], a closely related species, requires warm fluctuating temperatures for maximum germination (Nishimoto and McCarty 1997). The greater response to warm fluctuating temperatures may be the cause of both crowfootgrass and goosegrass emergence on bare ground (Adu et al. 1994; Gupta 1973; Nishimoto and McCarty 1997; Sharma and Chivinge 1982), where the greatest diurnal fluctuations would be expected.

### Response to Solution pH

ANOVA indicated a significant main effect of solution pH treatment, so crowfootgrass germination is presented by solution pH treatment averaged over temperature regimes (Figure 4). Crowfootgrass seed had the highest cumulative germination at solution pH of 4, and cumulative germination decreased with increasing pH. Cumulative seed germination and rate (*K*) was greater at solution pH 4 and 5 than at all other solution pHs, indicating that crowfootgrass germination is sensitive to changes in solution pH. Germination for each solution pH began within 2 to 6.2 d of exposure of seed to the treatment solution. Interestingly, Buchanan et al. (1975) observed that growth of crowfootgrass was less when grown in soil with pH 5.4 than at pH 6.3. However, in terms of yield relative to the treatment

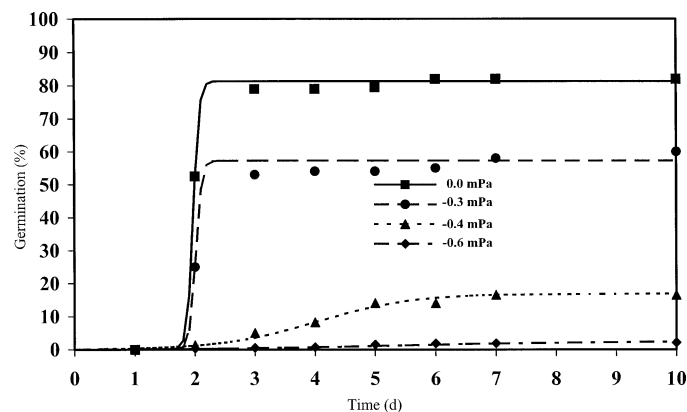


FIGURE 5. Influence of water stress on crowfootgrass germination averaged across temperature regimes, modeled using the equation  $y = M(1 + \exp[-K(t - L)])^{-1}$  with estimated parameters (and standard errors) as follows:

mPa	<i>M</i>	<i>K</i>	<i>L</i>	<i>R</i> <sup>2</sup>
0.0	81.3 (2.3)	19.8 (4.8)	2.0 (0.2)	0.75
-0.3	57.3 (3.3)	19.6 (0.3)	2.0 (0.3)	0.83
-0.4	16.9 (8.4)	1.3 (1.2)	4.1 (0.3)	0.69
-0.6	2.5 (0.5)	0.67 (0.6)	5.5 (1.5)	0.89

yielding the highest biomass, crowfootgrass produced more growth at lower pH than other species studied including redroot pigweed (*Amaranthus retroflexus* L.), prickly sida (*Sida spinosa* L.), and jimsonweed (*Datura stramonium* L.) (Buchanan et al. 1975). Together, these data suggest that crowfootgrass prefers acidic soil conditions, which are common throughout the major crop production regions of the North Carolina Piedmont and Coastal Plain (Tucker et al. 1997). Adaptation to acid soils is typical of the chloridoid grasses, of which crowfootgrass is a member (Surrey 1986).

### Response to Water Stress

ANOVA indicated a significant main effect of water stress treatment, thus crowfootgrass germination is presented by water stress treatment averaged over temperature regime (Figure 5). As water stress increased, cumulative crowfootgrass seed germination decreased. No germination occurred when the water potential was  $-0.8$  or  $-1.2$ , regardless of the germination temperature (data not shown). When the water potential was 0.0 (seed in deionized water), maximum germination was 85% averaged across the four temperature regimes. Placing seeds in water stress delayed the onset of germination, causing the time taken for 50% germination (*L*) to increase for  $-0.3$  mPa,  $-0.4$  mPa, and  $-0.6$  mPa compared with 0.0 mPa (Figure 5). The requirement for low water stress suggests that crowfootgrass is dependent upon a precipitation or an irrigation event for germination in the field.

### Depth of Emergence

Emergence of crowfootgrass decreased with increased planting depth, with the maximum of 40% occurring at 14 d after planting (DAP) from the 1-cm planting depth (Figure 6). At 7 DAP crowfootgrass emergence was greater from burial depths of 0.5 and 1 cm than any other depth of burial. Emergence was similar when seeds were planted on



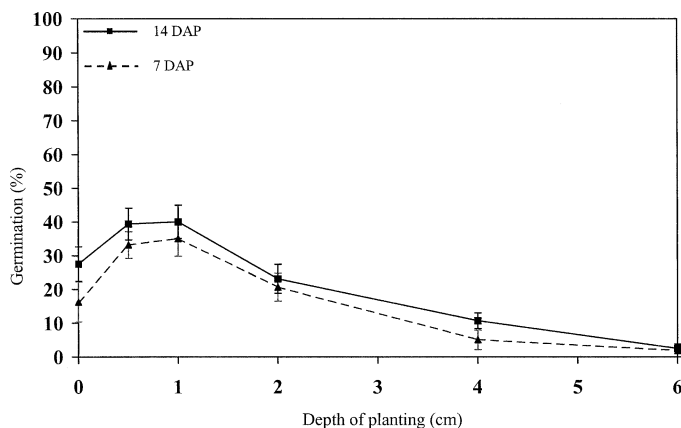


FIGURE 6. Cumulative emergence of crowfootgrass seed buried 0, 0.5, 1, 2, 4, or 6 cm 7 and 14 d after planting. Vertical bars represent standard errors of the mean.

the surface, at 0.5, or 1 cm deep, 14 DAP. Emergence on the surface or from a depth of 4 cm increased from 7 to 14 DAP. Delayed emergence from depths of 4 and 6 cm may be due to the larger distance to extend the coleoptile to the soil surface. Others have noted decreasing germination of crowfootgrass seed with increasing burial depth in populations from Nigeria and India, and the preponderance of germination occurred at burial depths of 1.5 cm or less (Kumar et al. 1971; Sharma and Chivinge 1982).

Larger seed with greater carbohydrate reserves can emerge from greater depths of burial (Baskin and Baskin 1998). Crowfootgrass seed is 0.7 to 1.0 mm in diameter. Broadleaf signalgrass possesses a larger seed, at 3 mm in length, as do other grasses such as giant foxtail (*Setaria faberi* Herrm.) (2 to 2.5 mm long) or fall panicum [*Panicum dichotomiflorum* (L.) Michx.] (2 to 2.5 mm long) (Hitchcock and Agnes 1971; Radford et al. 1973). Crowfootgrass percent emergence was similar to broadleaf signalgrass, fall panicum, and giant foxtail in glasshouse trials at depths of 0 and 1 cm and less than all three when buried deeper than 2 cm (Fau-sey and Renner 1997). Both fall panicum and giant foxtail germinated from a depth of 7.5 cm in glasshouse trials (Fau-sey and Renner 1997), whereas broadleaf signalgrass germinated to a depth of 6 cm, but no germination was observed from a depth of 10 cm (Burke et al. 2002). Only 3% of crowfootgrass seed emerged from a planting depth of 6 cm, 14 DAP (Figure 6).

Crowfootgrass did not tolerate water stress and required warm alternating temperatures, burial depths of 0 to 1 cm, and an acidic solution pH for maximum germination. These data suggest that crowfootgrass may emerge in the months of June, July, and August and germination is likely triggered by a precipitation event. Furthermore, if conditions are right, crowfootgrass will germinate rapidly. The response of crowfootgrass seed to fluctuating temperatures and shallow burial depths may indicate adaptation by crowfootgrass to disturbed bare ground (Adu et al. 1994; Gupta 1973; Nishimoto and McCarty 1997; Sharma and Chivinge 1982), where the greatest diurnal fluctuations would be expected. Shallow cultivation for weed control could potentially stimulate germination by placing crowfootgrass seed at the optimum depth for germination. The aforementioned attributes may also contribute to poor control later in the season by soil-applied herbicides as they degrade in the soil. Ad-

ditionally, high weed densities have been shown to decrease herbicide efficacy (Doub et al. 1988; Hartzler and Roth 1993). Emergence after final postemergence herbicide applications may also contribute to a lack of season-long control in many weed management programs (Prostko et al. 2001).

Other grass weeds with similar attributes (rapid germination), such as woolly cupgrass [*Eriochloa villosa* (Thunb.) Kunth] and giant foxtail have been shown to decline rapidly from the seedbank (Buhler and Hartzler 2001). A weed control system that took advantage of the seed depletion and controlled late season weed escapes might deplete the soil of crowfootgrass seed in several seasons. These attributes should be taken into account when managing crowfootgrass.

## Sources of Materials

<sup>1</sup> Seed Blower, Seedburo Equipment Company, 1022 W. Jackson Blvd., Chicago, IL 60607.

<sup>2</sup> SG8S Germinator, Hoffman Manufacturing Inc., International Agri-Supply, 353 29th Avenue SW, Albany, OR 97321.

<sup>3</sup> Watman #3 filter paper, Fisher Scientific, P.O. 4829, Norcross, GA 30091.

<sup>4</sup> 9.0 cm germination paper, Anchor Paper Company, 480 Broadway, St. Paul, MN 55165-0648.

<sup>5</sup> PEG 8000, Sigma Chemicals, P.O. Box 14508, St. Louis, MO 63178.

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